

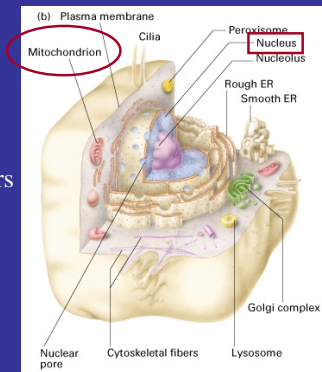
*The number of genes prescribing a eukaryotic life form such as a Douglas fir or human being runs into the tens of thousands. The nucleotide pairs composing them vary among species from one billion to ten billion. If the DNA helices in one cell of a mouse, a typical animal species, were placed end on end and magically enlarged to have the same width as wrapping string, they would extend for over nine hundred kilometers, with about four thousand nucleotide pairs packed into very meter. Measured in bits of pure information, the genome of a cell is comparable to all editions of the Encyclopedia Britannica published since its inception in 1768.*

**Table 3.1.** Historical overview of primary methods used to study genetic variation in natural populations.

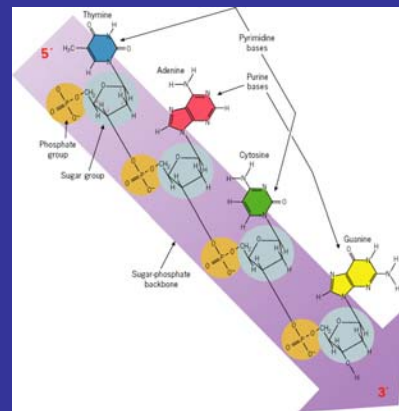
Time Period	Primary techniques
1900-1970	Laboratory matings and chromosomes
1970s	Protein electrophoresis (allozymes)
1980s	Mitochondrial DNA
1990s	Nuclear DNA
2000s	Genomics

## DNA

~17,000  
base pairs



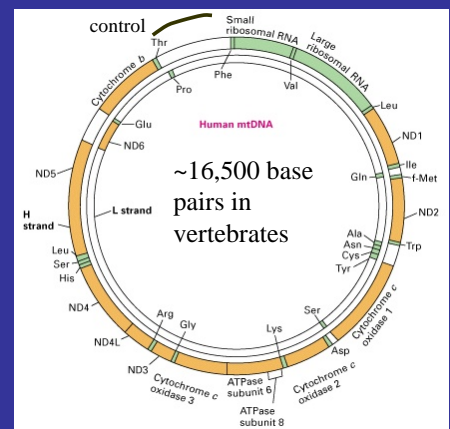
billions of  
base pairs



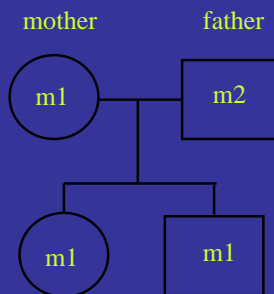
## Mitochondrial DNA (mtDNA)

- Predominantly **maternally inherited** in plants and animals
- **Haploid** (one copy in each individual)
- **No recombination**
- Location of oxidative phosphorylation (OXPHOS), which is vital for metabolic activity of somatic cells and gametes
- mtDNA encodes 13 of 67 components of the OXPHOS system (54 nuclear)

## mtDNA



## Maternal inheritance of mtDNA



All children inherit their mother's mtDNA type

## Mitochondrial genome variation and the origin of modern humans

Max Ingman\*, Henrik Kaessmann†, Svante Pääbo† & Ulf Gyllenstein\*

\* Department of Genetics and Pathology, Section of Medical Genetics, Rudbeck Laboratory, University of Uppsala, S-751 85 Uppsala, Sweden

† Max Planck Institute for Evolutionary Anthropology, Inselstrasse 22, D-04103 Leipzig, Germany

Compared 53 complete genomes (16,566 base pairs).  
2000. Nature 408:708-713.

## Within species polymorphism (humans)

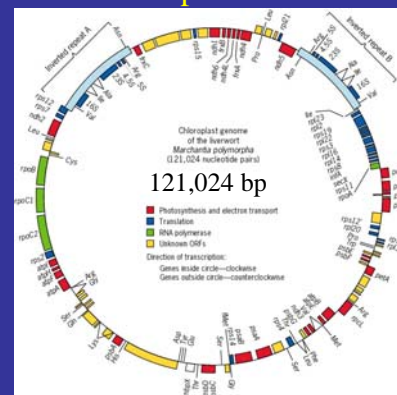


Asians & others

Asians

Africans

## Chloroplast DNA



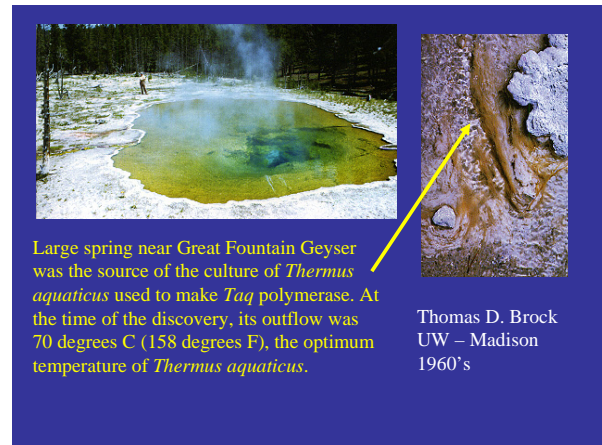
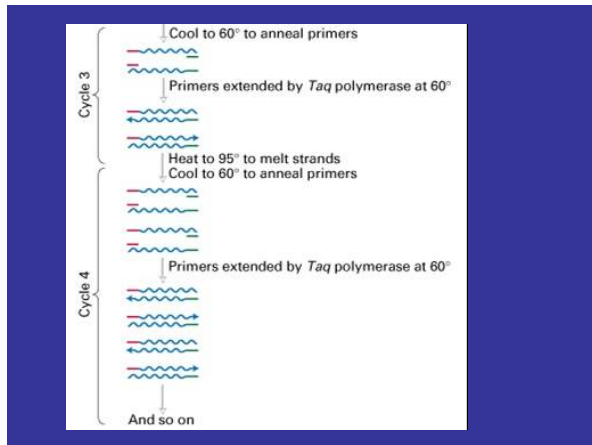
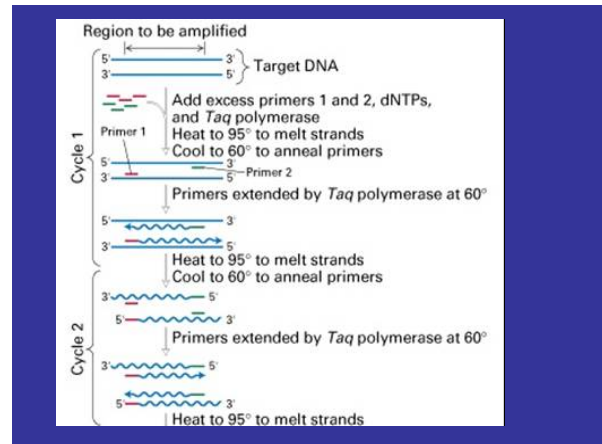
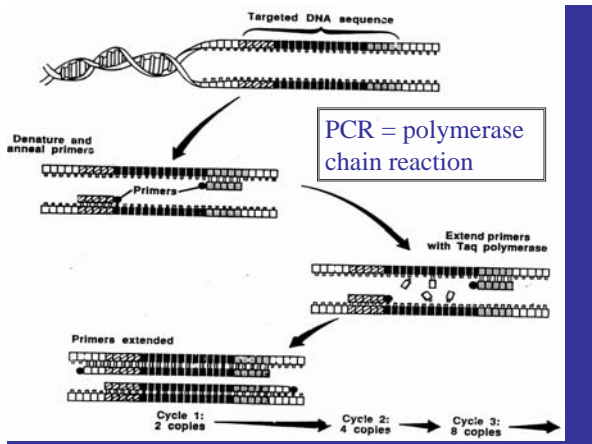
Liverwort  
*Marchantia polymorpha*

## Chloroplasts

- Location of photosynthesis in plant cells
- **Haploid** (one copy in each individual)
- **Maternally** inherited in some groups and **paternally** inherited in others (pine trees)

## Polymerase Chain Reaction (PCR)

- Allows us to make many copies of a specific region of the genome (amplification)
- We need two primers that flank a certain gene or region in the species of interest
- Primers are short (~20 bp) synthetic strands of DNA that bind to DNA and start the replication process



## Which Whales Are Hunted? A Molecular Genetic Approach to Monitoring Whaling

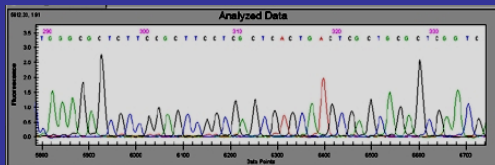
C. S. Baker and S. R. Palumbi

Minke whale

Tokyo fish market

Hotel PCR

## Mitochondrial DNA Sequencing

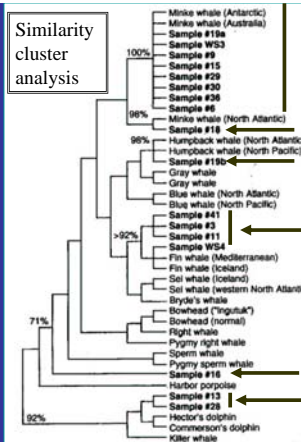


...CTGAAAGGGT... Haplotype A  
...CTGAAAGGGT... Haplotype B



Base substitution

Similarity  
cluster  
analysis



Southern minke (8)

Northern minke

Humpback

Fin whale (4)

Unknown "whale"

Unknown "dolphins" (2)

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Time Period	Primary techniques
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Andrea Taylor and friend



Microsatellite loci

or

VNTR (Variable  
Number of Tandem  
Repeat) loci

or

SSR (Simple sequence  
repeats)

or

STRPs (short random  
repeat polymorphisms)

**Box 3.2.** Microsatellite DNA sequence polymorphism.

Below is a locus on a chromosome showing two alleles from an individual heterozygous for a microsatellite dinucleotide repeat (AC; in bold and blue). Note the difference in allele length results from an addition of one repeat in the lower (paternal) allele. The primer-binding sites for PCR are underlined. They are highly conserved sites (usually ~20 bp long) flanking the highly variable microsatellite repeats.

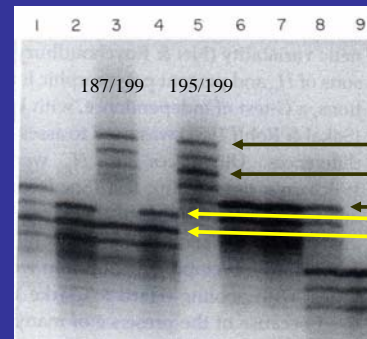
Maternal chromosome (allele with 7 repeats):

primer site  
5' - TGACTTAGTGTATACATACACACACACACACTTATCGATCGAGCTAT - 3'  
3' - ACTGAATCACATATGTATGTGTGTGTGTGAATAGCTAGCTCGATA - 5'  
primer site

Paternal chromosome (allele with 8 repeats):

primer site  
5' - TGACTTAGTGTATACATACACACACACACACTTATCGATCGAGCTAT - 3'  
3' - ACTGAATCACATATGTATGTGTGTGTGTGAATAGCTAGCTCGATA - 5'  
primer site

## Hairy-nosed wombat microsatellite



Base pairs

199

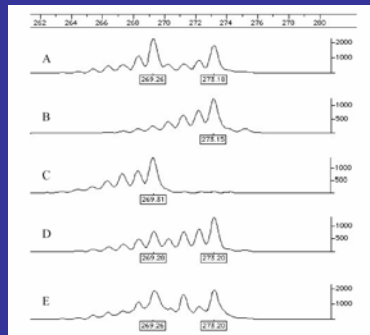
195

191

189

187

183



## Genotypes

269/273

273/273

269/269

269/273

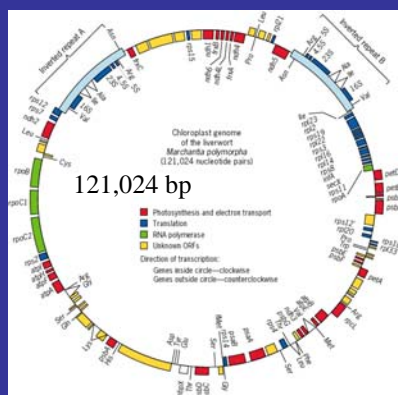
269/273 (?)

European Alpine ibex (*Capra ibex*)

Box 4.3. Modified GenBank sequence database entry for the *Lla71CA* locus in the hairy-nosed wombat (Figure 4.4). The primers in the sequence at the bottom have been capitalized and the repeat region (CA) is shown in bold. The n's in the sequence are basepairs that could not be resolved in the sequencing process.

```
1: AF185107. Lasiorchinus latif
LOCUS       AF185107             310 bp    DNA    linear    MM 01-JAN-2000
DEFINITION  Lasiorchinus latifrons microsatellite Lla71CA sequence.
AUTHORS    Beheregaray,L.B., Sunnucks,P., Alpers,D.L. and Taylor,A.C.
TITLE       Microsatellite loci for the hairy-nosed wombats (Lasiorchinus
            krefftii and Lasiorchinus latifrons)
JOURNAL     Unpublished
AUTHORS     Taylor,A.C.
JOURNAL     Submitted (31-AUG-1999) Biological Sciences, Monash University,
            Wellington Rd., Clayton, VIC 3168, Australia
FEATURES             Location/Qualifiers
     source         1..310
     repeat_region   109..154
                   /rpt_type=tandem
                   /rpt_unit=ca
BASE COUNT   99 a    94 c    42 g    68 t    7 others
ORIGIN        1  gngctggggn  ccocctgggato  acagaatota  aatctgagca  tctcagaAATG  AGAAGGTATC
              61  TCCAGGataa  ccannncocot  ctactaaac  agaattoca  ctccotaca  cacacacaca
              121  cacacacaca  cacacacaca  cacacacaca  cacactaat  agacacaca  agtggaaagt
              181  cacacagcct  ttggggnagp  tgggggatat  acttCCTATG  ACATAGCCTA  TACCaactot
              241  gaatagtaac  ttctactatc  ataataotaa  aactactctc  ccactctttt  ctgtagtttc
              301  tataatctgg
```

## Microsatellites in Chloroplast DNA!!!



## Evolutionary Conservation of Ten Microsatellite Loci in Four Species of Felidae

M. A. Menotti-Raymond and S. J. O'Brien Journal of Heredity 86:319-322. 1995.

Heterologous **microsatellite primers**

**Fca 8:** ACTGTAAATTTCTGAGCTGGCC  
TGACAGACTGTTCTGGGTATGG

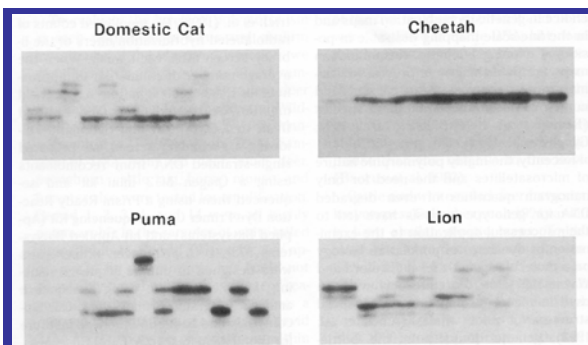
**Fca 23:** CAGTTCCTTTTCTCAAGATTGC  
GCAACTCTTAATCAAGATTCATT

**Fca 35:** CTTGCCTCTGAAAAATGTAAATG  
AAACGTAGGTGGGGTTAGTGG

**Fca 43:** GAGCCACCCTAGCACATATACC  
AGACGGGATTGCATGAAAAG

**Fca 45:** TGAAGAAAAGAATCAGGCTGTG  
GTATGAGCATCTCTGTGTTCTGTG

Domestic cat primers



Fca77



Is there mtDNA sequence information for your favorite species?

[GenBank](#)

National Center for Biotechnology Information (NCBI)

### Individual ID

Through the most sensitive DNA testing, RFLP testing, the FBI Laboratory determined conclusively that the semen on Ms. Lewinsky's dress was, in fact, the President's. The chance that the semen is not the President's is one in 7.87 trillion.

$$\text{Prob} = 1 / (7,870,000,000)$$

### Microsatellites as genetic tags

(mark-recapture)



Dry Fork Valley

Two Medicine River



Rub tree



### Ancient DNA

Molecular Ecology 1997, 6, 487-492

#### SHORT COMMUNICATION

### Analysis of microsatellite DNA from old scale samples of Atlantic salmon *Salmo salar*: a comparison of genetic composition over 60 years

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†Danish Institute for Fisheries Research, Department of Inland Fisheries, Vejlsøvej 39, 8600 Silkeborg, Denmark

#### Abstract

Microsatellite analysis was applied to scale samples of Atlantic salmon collected up to 60 years ago. Samples from the 1930s, from a now endangered Danish population, were compared with recent samples (1989), to test if the present population consists of descendants from the original one. Allele frequencies had changed over time, but individuals from the two samples caught about 60 years apart clustered together when compared with the closest neighbouring population and another reference population. However, fewer alleles were detected in the recent sample from the endangered population, most likely due to a population bottleneck or sampling artefacts.

Locus	Allele	Skjern River		Conon	Åtran
		1989	1930s		
SSOSL 85	177		0.007		
	179		0.010		
	181		0.007		
	183	0.11	0.316	0.08	0.15
	187		0.003		
	189		0.003	0.46	0.14
	191		0.003	0.02	0.02
	193	0.06	0.076	0.08	0.25
	195	0.40	0.330	0.02	0.02
	197		0.017	0.04	0.13
	199	0.28	0.073	0.21	0.11
	201	0.14	0.142	0.02	0.06
	203			0.08	0.04
	209				0.02
	217		0.003		0.04
	221	0.01	0.007		0.02